



## Identifying the most important traits affecting grain yield of wild wheat (*Triticum boeoticum*) under drought stress conditions

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### ABSTRACT

Boeoticum specie is a valuable drought-tolerance gene source to breed wheat yield under stress. This study was done to identify the most important traits affecting grain yield of 10 boeoticum ecotypes under drought stress conditions for two years. Water use efficiency, fertile spikes number per plant and seed number per plant showed the highest positive and significant ( $p \leq 0.01$ ) correlation with grain yield per plant. Water use efficiency, fertile spikes number per plant, seed number per the main spike, biological yield per plant, and water use (with a negative regression coefficient), as the most important traits, were entered into the regression model, respectively. The most direct effect on increasing grain yield was water use efficiency. Seed number per plant and fertile spikes number per plant, due to increased water use efficiency, showed the most indirect effect on grain yield. Ecotype 5, as a drought-tolerant ecotype, showed a high water use efficiency by allocating more assimilates to yield components. It had a high grain yield. On the other hand, ecotype 6 was introduced as the most drought-susceptible ecotype with low-economical yield. In this study, high water use efficiency increased the traits related to seed number per plant. The ratio of assimilating allocation to aboveground or underground parts was the main mechanism for the adaptation of ecotypes. Therefore, selection based on these mechanisms will lead to the identification of drought-tolerant ecotypes for future wheat breeding programs.

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## Introduction

Wheat is the most important grain in the world and in Iran, provides about 20% of the needed calories for people. Due to population growth and climate changes in recent years, the demand for this strategic plant is increasing day by day (Akpinar, *et al.*, 2013). Among environmental stresses, drought is the most important stress that significantly reduces the annual cultivation area, growth rate, and yield of this important crop (Moosavi *et al.*, 2020). Iran is an arid and semi-arid region of the world with an average annual rainfall of 240 mm. Due to the lack of

access to water resources in rain-fed areas, an evaluation of the effects of this stress on the growth and development of wild wheat ancestral plants, as a genetic source of drought resistance, is necessary (Naderi and Eslahi, 2019). Despite the growing demand for this important crop, a significant decline in the yield of this plant has been observed due to drought. About 70% of the world's arable land is directly and indirectly affected by the adverse effects of this stress (Budak *et al.*, 2013). With the closure of the stomata under drought stress, the uptake of carbon dioxide decreases, and consequently, the rate of photosynthesis decreases (Khaleghi *et al.*, 2014). Depending on the duration and severity of drought stress, the amount of damage to the plant will vary. For example, in conditions of very severe drought stress, not only the plant yield is reduced, but also the plant may be completely damaged and destroyed (Akpınar, *et al.*, 2013). Given that, the majority of wheat fields are always affected by drought stress, the production and development of drought-tolerant cultivars are one of the most important goals of breeding programs in most parts of the world (Budak *et al.*, 2013). Therefore, identifying the root characters, morpho-physiological traits, and phenological characteristics responding to drought stress in wheat progeny and selecting cultivars based on these traits or transferring them to cultivars is a common and desirable solution (Liu *et al.*, 2006). Wild relatives of wheat are considered one of the valuable genetic sources for breeding drought tolerance in wheat (Nazari *et al.*, 2019). Among the wild relatives of wheat, i.e. *T. boeoticum*, *T. monococum*, and *T. urartu*, the species of *boeoticum* is more tolerant to drought stress (Munns *et al.*, 2012). According to previous research, different ecotypes of *boeoticum* species are resistant to drought stress (Pour-Aboughadareh *et al.*, 2016), salinity stress (Munns *et al.*, 2012), and pathogens (6). Therefore, different ecotypes of this species are used as a valuable source of genes for wheat breeding.

Typically, different statistical methods such as correlation analysis, regression analysis, path analysis, and principal component analysis are used to evaluate and interpret the relationships between different traits (Mohammadi and Prasanna, 2003). Using these methods, the relationships between different traits with grain yield can be evaluated to identify traits affecting grain yield and then determine the desired genotypes. By identifying and selecting to improve grain yield-related traits, it is possible to improve yield indirectly (Blum *et al.*, 2011). Although the correlation coefficient shows linear relationships between traits, estimating the correlation coefficient between yield and its components is not enough to achieve the effect and importance of these components on grain yield (Ali and Shakor, 2012). In previous research (Munns *et al.*, 2012 and Passioura, 1996) different phenological, morphological, physiological and root traits have been used to select drought-tolerant genotypes in wheat.

The main purpose of this study was to evaluate the agro-morpho-physiological traits of 10 ecotypes of *boeoticum* specie under drought stress. Therefore, the best ecotypes and the most important traits affecting the increase of grain yield under drought conditions were identified for using wheat breeding programs in the future.

## Material and methods

To evaluate the root traits, this study was carried out in a research greenhouse of Bu-Ali Sina University in Hamedan, under drought stress conditions of 45% field capacity, according to Nazari *et al.*, (2018). Plant materials included 10 ecotypes of *Triticum boeoticum*, belonging to different regions of Iran (Table 1). Ecotypes were evaluated in a randomized complete block design with three replications. Each pot had a height of 40 cm and a diameter of 30 cm, which was filled with 15 kg of pot soil, with a combination of 50% field soil, 25% aeolian sand, and 25% rotted manure. Initially, 15 grains were planted in each pot, but after germination, the seedlings were decreased to 10 in each pot. Up to three weeks after planting, the moisture of the pots was maintained at 100% of the field capacity. After establishment, water stress

treatment was applied at 45% of field capacity in the 4-6 leaf stage (Moosavi *et al.*, 2017; Nazari *et al.*, 2018). Drought stress treatment continued until the last stage of growth, i.e. until the stage of physiological maturity.

To calculate the exact weight of the pots without the weight of the plants inside them, some pots were planted in addition to the pots under test.

After starting the drought stress treatment, the plants related to these extra pots were removed and weighed. The average weight of additional plants at each stage before watering was subtracted from the weight of the pots. Finally, the exact amount of water required to reach 45% of the field capacity was determined.

**Table 1.** The studied genotypes under drought stress conditions for two years

Accession code	Scientific name	Site of collection (city, province, country)
Tb <sub>1</sub>	<i>Triticum boeoticum</i>	Dehsefid, Kermanshah, Iran.
Tb <sub>2</sub>	<i>Triticum boeoticum</i>	Khoramabad road to Firouzabad, Lorestan, Iran.
Tb <sub>3</sub>	<i>Triticum boeoticum</i>	150 Km before Mianeh from Ardabil, Iran.
Tb <sub>4</sub>	<i>Triticum boeoticum</i>	Ravansar, Kermanshah, Iran.
Tb <sub>5</sub>	<i>Triticum boeoticum</i>	1 Km before Zagheh from Droud, Lorestan, Iran.
Tb <sub>6</sub>	<i>Triticum boeoticum</i>	20 Km after Sarvabad, road of Sanandaj to Marivan, Kurdistan, Iran.
Tb <sub>7</sub>	<i>Triticum boeoticum</i>	10 Km before Norabad from Aleshtar, Lorestan, Iran.
Tb <sub>8</sub>	<i>Triticum boeoticum</i>	Ahar, East-Azarbaijan, Iran.
Tb <sub>9</sub>	<i>Triticum boeoticum</i>	10 Km after Ganji to Ghorveh, Kurdistan, Iran.
Tb <sub>10</sub>	<i>Triticum boeoticum</i>	Ghorveh, Kurdistan, Iran.

**Table 2.** The information and abbreviations of 32 measured traits during two years

Character	Abbreviation	Character	Abbreviation
Days to heading	DTH	Main stem weight (g)	MSTW
Days to anthesis	DTA	1000-grain weight (g)	TKW
Days to maturity	DTM	Economical yield per plant (g)	EYPP
Grain filling period	GFP	Biological yield per plant (g)	BYPP (SDW)
Chlorophyll content (%)	SPAD	Plant harvest index (%)	PHI
Plant height (cm)	PH	Leaf area index (cm <sup>2</sup> )	LAI
Peduncle length (cm)	PEL	Relative water content (%)	RWC
Leaves number	LN	Excised leaf water retention (%)	ELWR
Tillers number per plant	TN	Water use (l)	WU
Fertile spikes number per plant	NFS	Water use efficiency (g/l)	WUE
Spikelet number per spike	SNPS	Main root length (cm)	MRL
Seed number per the main spike	SNPMS	Main root volume (cm <sup>3</sup> )	MRV
Seed number per plant	SNPP	Root dry weight (g)	RDW
Main spike weight (g)	MSPW	Root area (cm <sup>2</sup> )	RA
Seed weight per the main spike (g)	SWPMS	Root-to-shoot dry weight ratio	RDWSDW
Peduncle weight (g)	PEW	Root diameter (cm)	RD

In this study, the relationship between 32 different traits (phenological, agro- morphological, physiological, and root traits) was evaluated to identify the most desirable traits affecting the grain yield (Table 2). The trait of the grain filling period was obtained from the difference between "the number of days to pollination" and "the number of days to physiological maturity". The relative water content of the leaves and excised leaf water retention were calculated according to the method of Magis *et al.* (2013). During the experiment, for each pot at each stage of irrigation, the amount of water used was recorded, and based on that, the amount of water consumption in each pot was obtained as the total amount of water used during plant growth. To measure the root traits, at first, the plants were removed from the soil surface of the pot, after washing the roots, root-related traits were measured. Water use efficiency in different experimental treatments was determined by calculating the ratio of grain yield to the amount of pure water consumed per plant (Vafabakhsh *et al.*, 2009). To calculate the net water consumption of the plant, five pots were brought to the desired moisture level at 100% of the

field capacity. Then, when draining the soil moisture to 45% of the pot capacity, re-irrigation was done up to 100% of the field capacity. This volume of water consumption showed the rate of evaporation from the surface of each pot, which, with its difference from the total water consumption, the amount of pure water consumed by the plant was obtained.

To measure the leaf area, after separating the leaves of each plant from the stem, the length (L) and the middle width of each leaf (W) were measured and then the leaf area (A) was calculated through the following equation (Moll and Kamparth, 1977);

$$A = L \times w \times 0.75 \quad (1)$$

The mean data of two years was used to evaluate the relationships between traits, correlation analysis, stepwise regression analysis in the forward method, causal analysis, and principal component analysis by Minitab software.

## Results

### *Correlation analysis results*

Water use efficiency, fertile spikes number per plant, seed number per plant, biological yield per plant, seed number per the main spike, main spike weight, and plant height showed a positive and significant ( $p \leq 0.05$ ) correlation with grain yield per plant, respectively (Table 3).

### *Stepwise regression results*

The results showed that water use efficiency, fertile spikes number per plant, seed number per the main spike, biological yield per plant, and water use (with negative regression coefficient) were entered into the regression model, respectively (Table 4). Water use efficiency, as the most important trait affecting grain yield increase under water stress conditions, alone explained 72.35% of yield changes and had the most positive effect on increasing grain yield.

### *Path analysis results*

The results (Table 5) showed that the traits of water use efficiency (0.987), fertile spikes number per plant (0.608), and seed number per plant (0.587), had the most direct effects on economical yield per plant under drought stress conditions, respectively.

The most indirect effect on increasing economical yield per plant was related to the seed number per plant (0.53) and fertile spikes number per plant (0.495), respectively. So that both traits, by directly increasing water use efficiency, will increase grain yield per plant in the desired genotypes (Table 5). A noteworthy point in the obtained results (Table 5) is the existence of a negative coefficient of direct and indirect effects of water use (-0.186 and -0.239, respectively) under water stress conditions.

### *Principal component analysis results*

The biplot results showed that the traits and ecotypes located in area A were the most desirable traits and ecotypes (ecotypes 5 and 3, respectively) (Figure 1). Conversely, the traits and ecotype (No. 6) located in area B were the most unfavorable traits and ecotypes for drought stress conditions. The results of the principal component analysis were in good agreement with the results of other statistical analyzes. Among the studied ecotypes, ecotypes 5 and 3 were the most tolerant and ecotype 6 was the most susceptible ecotype to drought stress (Figure 4.).

**Table 3.** Correlation between different traits measured in 10 wild wheat ecotypes (*Triticum boeoticum*) during two years

Trait	DTH	DTA	DTM	GFP	PH	PEL	TN	LN	SPAD	LAI	RWC	ELWR	MSPW	SPNPS	SWPS	SNPS
DTA	0.97**															
DTM	0.27	0.35														
GFP	-0.89**	-0.86**	0.13													
PH	-0.31	-0.14	-0.21	0.08												
PEL	-0.40	-0.48*	-0.46*	0.31	0.80**											
TN	0.38	0.42	0.24	-0.30	-0.14	-0.31										
LN	0.35	0.39	0.33	-0.23	-0.20	-0.37	0.98**									
SPAD	-0.28	-0.30	-0.40	0.11	0.22	0.43	0.31	0.28								
LAI	-0.27	0.32	-0.04	-0.32	0.01	0.01	0.46*	0.44	0.15							
RWC	-0.48*	-0.34	0.14	0.44	-0.40	-0.27	0.08	0.14	0.01	0.03						
ELWR	0.02	-0.06	-0.07	0.05	-0.09	-0.18	0.16	0.17	-0.10	0.15	-0.13					
MSPW	0.01	0.01	0.06	0.08	0.63**	0.53*	0.22	0.21	0.20	0.20	-0.01	-0.13				
SPNPS	0.52*	0.51*	0.07	-0.47*	0.19	-0.05	0.81**	0.76**	0.37	0.44*	-0.14	0.12	0.37			
SWPS	-0.12	-0.09	0.16	0.24	0.52*	0.50*	0.12	0.12	0.15	0.14	0.14	-0.24	0.96**	0.20		
SNPS	0.49*	0.52*	0.23	-0.40	0.07	-0.10	0.91**	0.89**	0.30	0.43	-0.06	-0.03	0.45*	0.84**	0.33	
MSTW	0.03	0.08	0.23	0.08	0.39	0.32	0.12	0.12	-0.01	0.36	0.33	-0.17	0.77**	0.22	0.81**	0.29
PEW	0.02	0.03	0.33	0.17	0.22	0.23	0.08	0.18	0.18	0.24	0.07	-0.17	0.58*	0.08	0.65**	0.22
NFS	0.34	0.35	0.47*	-0.17	-0.14	0.38	0.34	0.42	-0.13	-0.14	-0.15	-0.05	-0.08	0.24	-0.13	0.38
BYPP	0.39	0.45*	0.43	-0.28	-0.19	-0.39	0.82**	0.84**	0.11	0.30	0.23	-0.08	0.23	0.65**	0.18	0.81**
EYPP	0.01	0.02	0.01	0.03	0.43*	0.27	0.34	0.34	0.15	-0.01	-0.11	-0.05	0.48*	0.36	0.37	0.49*
TKW	-0.52*	-0.49*	-0.22	0.45*	0.45*	0.62**	-0.43	-0.44	0.11	0.05	0.20	-0.24	0.59**	-0.28	0.68**	-0.31
SNPP	0.47*	0.48*	0.38	-0.31	-0.04	-0.30	0.81**	0.84**	0.16	0.21	-0.19	0.05	0.12	0.71**	0.03	0.81**
MRL	-0.18	-0.02	0.44	0.25	-0.60**	-0.44	-0.12	-0.06	-0.34	-0.13	0.62**	-0.31	-0.19	-0.037	0.03	-0.20
MRV	0.43	0.52*	0.70**	-0.18	-0.13	-0.37	0.47*	0.53*	-0.26	0.29	0.22	-0.12	0.35	0.34	0.39	0.55*
WU	0.29	0.33	0.49*	-0.17	-0.39	-0.50*	-0.17	-0.08	-0.31	-0.20	0.02	-0.25	-0.32	-0.24	-0.22	-0.17
PHI	-0.34	0.42	-0.59**	0.17	0.38	0.59**	-0.37	-0.42	0.23	-0.11	-0.45*	0.11	0.04	-0.27	0.06	-0.33
WUE	0.01	-0.04	-0.05	0.03	0.49*	0.35	0.34	0.35	0.21	0.04	-0.18	-0.02	0.52*	0.39	0.40	0.50*
AR	0.25	0.40	0.70**	-0.06	-0.36	-0.47*	0.30	0.35	-0.42	0.20	0.49*	-0.24	0.20	0.11	0.32	0.33
RD	0.11	0.23	0.60**	0.04	-0.31	-0.40	0.30	0.37	-0.12	0.15	0.53*	-0.12	0.24	0.14	0.34	0.31
DRWBY	0.55*	0.56*	0.18	-0.47*	-0.21	-0.33	0.85**	0.85**	0.29	0.52*	-0.20	0.26	0.14	0.77**	0.01	0.81**
RDW	-0.09	-0.07	0.55**	0.21	-0.34	-0.27	0.17	0.23	-0.15	0.20	0.67**	-0.35	0.25	-0.01	0.43	0.17

**Table 3.** Continued

Trait	MSTW	PEW	NFS	BYPP	EYPP	TKW	SNPP	MRL	MRV	WU	PHI	WUE	AR	RD	DRWBY
PEW	0.62**														
NFS	-0.23	0.10													
BYPP	0.26	0.23	0.60**												
EYPP	0.16	0.14	0.60**	0.50*											
TKW	0.50*	0.32	-0.52*	-0.43	0.03										
SNPP	-0.07	0.13	0.76**	0.80**	0.58**	-0.56**									
MRL	0.15	0.02	-0.22	-0.01	-0.56*	0.10	-0.32								
MRV	0.55*	0.54*	0.46*	0.69**	0.24	-0.13	0.50*	0.24							
WU	-0.11	0.31	0.48*	0.19	-0.18	-0.33	0.10	0.27	0.41						
PHI	-0.33	-0.18	-0.21	-0.62**	0.15	0.40*	-0.27	-0.45*	-0.66**	-0.42					
WUE	0.16	0.18	0.54*	0.45*	0.98**	0.07	0.58**	-0.63**	0.18	-0.26	0.24				
AR	0.52*	0.31	0.20	0.51*	-0.05	-0.01	0.20	0.65**	0.86**	0.36	-0.69**	-0.13			
RD	0.47*	0.46*	0.36	0.60**	0.14	-0.01	0.24	0.38	0.84**	0.46*	-0.69**	0.05	0.81**		
DRWBY	-0.20	0.18	0.32	0.64**	0.16	-0.46*	0.75**	-0.21	0.43*	-0.04	-0.31	0.20	0.19	0.25	
RDW	0.57*	0.41*	-0.04	0.38	-0.12	0.26	-0.13	0.75**	0.68**	0.26	-0.59**	-0.20	0.89**	0.80**	0.05

Abbreviations of the traits are listed in Table 2

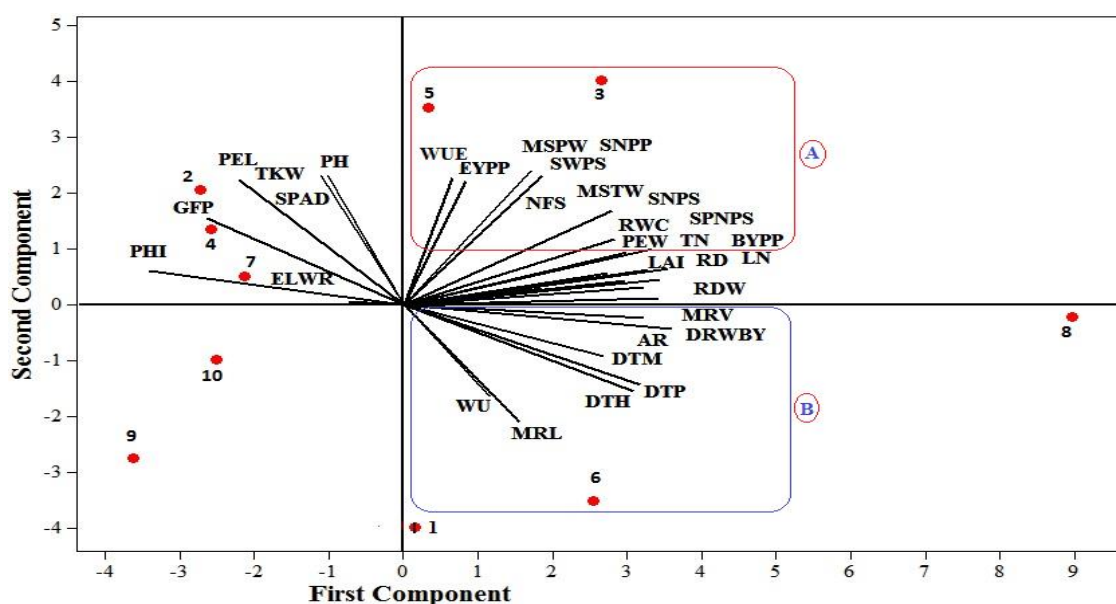
**Table 4.** Stepwise regression results for yield as a dependent variable in 10 wild wheat ecotypes (*Triticum boeoticum*) studied for two years

Trait	Intercept	Regression coefficients of traits in different steps of regression analysis					Cumulative detection coefficient
		1	2	3	4	5	
Water use efficiency	0.482	19.0		72.35**			
Fertile spikes number per plant	0.020	18.0	0.01	83.09**			
Seed number per plant	0.007	18.3	0.02	91.25**			
Biological yield per plant	-0.012	18.4	0.02	97.23**	0.017		
Water use	0.170	17.5	0.03	99.30*	0.019	-0.002	

**Table 5.** Direct and indirect effects of traits in grain yield regression model on 10 wild wheat ecotypes (*Triticum boeoticum*) studied for two years

Trait	The direct effect of trait on grain yield	Indirect effects on grain yield through other traits					Correlation with grain yield
		1	2	3	4	5	
Water use efficiency	0.987	-----	0.129	-0.155	0.082	0.018	0.987**
Fertile spikes number per plant	0.608	0.497	-----	-0.204	0.110	-0.034	0.608**
Seed number per plant	0.587	0.530	0.181	-----	0.147	-0.008	0.587**
Biological yield per plant	0.506	0.407	0.142	-0.214	-----	-0.014	0.506*
Water use	-0.186	-0.239	0.114	-0.029	0.036	-----	-0.186 <sup>ns</sup>

Residual effect= 0.077



**Figure 1.** Biplot graph of the first and second main components for 32 traits studied in 10 wild wheat ecotypes (*Triticum boeoticum*) over two years (traits abbreviations are listed in Table 2). A; Desirable biplot area, B; Undesirable biplot area

## Discussion

According to the results (Table 3), the highest positive correlation coefficient was observed between grain yield and water use efficiency. Passiora (1996) stated the three traits of water consumption, water use efficiency, and harvest index as three factors determining grain yield under drought stress. Therefore, according to previous studies (Lopez *et al.*, 2017; Nazari *et al.*, 2018), water use efficiency can be suggested as a valuable indicator for indirect selection to improve grain yield under limited moisture conditions. According to the results (Table 3), increasing the number of traits that will increase the number of grains per plant is an effective factor to improve grain yield per plant in the studied germplasm. Therefore, excessive allocation of assimilates to underground parts, for example increasing the length of the main root, leads to a decrease in the allocation of assimilates over ground reproductive parts, so it will reduce grain yield per plant. The correlation between grain yield and main root length was negative and significant ( $r = -0.56^{**}$ ). In a study on *Aegilops tauschii* under water stress (Moosavi *et al.*, 2017), it was stated that water use efficiency is an important and effective trait. It had a positive and significant correlation with grain yield per plant. Even though grain yield was associated with the traits of seed number per plant ( $r = 0.58^{**}$ ), fertile spikes number per plant ( $r = 0.60^{**}$ ), and seed number per the main spike ( $r = 0.49^{**}$ ), grain yield showed a weak correlation with 1000-grain weight ( $r = 0.03$ ). According to these results, it can be inferred that by increasing the number of grains per plant, the competition between florets for photosynthetic material has increased, leading to a decrease in 1000-grain weight and ultimately a weak relationship between this trait and grain yield. In addition, a negative and significant correlation between the number of grains per plant and 1000-grain weight ( $r = -0.56^{**}$ ) also confirms this object (Table 3). This result was consistent with the previous studies on wheat (Dewey and Lu, 1959; Mohammad, 2014). As shown in Table 3, spike-related traits such as grain weight per the main spike, the number of grains per main spike, and the number of spikelets per spike had a positive and significant correlation with each other (Moghaddam *et al.*, 2012).

Given the negative relationship between some root traits and grain yield of wheat (Leilah and Al-Khateeb, 2005) and wild wheat (Naghavi and Amirian, 2005) under limited moisture conditions, it can be stated that ecotypes are more successful in improving yield than a limited

part of their assimilates allocate to underground sections and allocate most of these materials more rapidly and in proportion to the economical or reproductive parts. In a study on wheat under water stress (Saba *et al.*, 2018), it was stated that grain yield had a positive and significant correlation with biomass traits per plant, the number of spikes per plant, 1000-grain weight, and grain filling period. Also, plant height, day to physiological maturity, root traits, and canopy temperature showed a negative correlation with grain yield. In general, according to the aim of this study, by indirect selection for high heritability traits that have a significant correlation with yield, it is possible to control environmental effects and achieve higher genetic gain and greater response to selection (Dawari and Luthra, 1991).

The results (Table 4) showed that water use efficiency, fertile spikes number per plant, seed number per the main spike, biological yield per plant, and water use (with negative regression coefficient), respectively, were entered into the grain yield regression model. These traits were the most important traits affecting grain yield changes under water stress. Therefore, diversity in these traits will lead to differences between ecotypes in terms of grain yield. Water use efficiency, as the most important trait affecting grain yield increase under water stress conditions, alone explained 72.35% of yield changes and had the most positive effect on grain yield increase. The results of regression analysis were in accordance with the results of correlation analysis, which is a confirmation of the accuracy of the results of these two statistical methods. The second group of important traits that were included in the regression model was the traits related to the number of seeds, as important components of grain yield, i.e. the fertile spike number per plant and the seed number per plant. This result indicates that the increase in grain yield in the optimal genotypes of this germplasm, mainly through increasing the number of seeds per plant and the weight of 1000 grains did not play an important role in increasing grain yield. In a study (Mojtabaie- Zamani *et al.*, 2014), it was stated that under heat stress conditions, only high grain filling speed during grain filling period cannot increase grain yield in wheat but also other factors such as grain filling period duration and the number of grains in spikes, play an important role in increasing yield under these stress conditions. However, in a study (Sayyah *et al.*, 2010), harvest index traits, 1000-grain weight, spike density, number of spikelets per spike, spike length, awn length, and grain filling period, as the most important traits affecting the grain yield of bread wheat were identified. In another study (Moosavi *et al.*, 2013), grain harvest index, plant biomass, and relative leaf water content were suggested as the most important traits affecting grain yield, which explained a total of 94% of yield variance.

Harb *et al.* (2012) stated that plant biomass, grain number per spike, spike number, and grain weight per spike had the most positive effect on the grain yield of native wheat masses. In our study, water use efficiency, fertile spikes number per plant and seed number per plant had a significant role in increasing grain yield in the studied germplasm and are suggested as desirable traits with a high response to selection.

Stepwise regression analysis is one of the most useful and practical statistical methods to identify the most important variables affecting a dependent variable, including grain yield (Mansourfar *et al.*, 2013). In other words, this method is used as a prerequisite method to introduce the best variables for path analysis.

The results (Table 5) showed that the traits of water use efficiency (0.987), fertile spikes number per plant (0.608), and seed number per plant (0.587), respectively, had the most direct effects on grain yield under drought stress conditions. The noteworthy point for these traits is that their correlation coefficient with grain yield is approximately equal to their direct effect coefficients with yield. This result implies that the selection of optimal ecotypes based on the high amount of these three traits will lead to an indirect increase in grain yield.

The most indirect effect on increasing grain yield was related to the seed number per plant (0.53) and fertile spike number per plant (0.495), respectively. So that both traits, by directly increasing water use efficiency, will increase grain yield in the desired genotypes (Table 5). A



noteworthy point in the obtained results (Table 5) is the existence of a negative coefficient of direct and indirect effects of water use (-0.186 and -0.239, respectively) under water stress conditions. The direct effect of this trait is through reduced grain yield and its indirect effect is through reduced water use efficiency. Indeed, although unfavorable ecotypes have high water consumption in drought stress conditions, their water use efficiency is not high, which is not favorable for limited moisture conditions. In previous studies on wheat (Dencic *et al.*, 2000; Solomon and Labuschagne, 2004), it was stated that fertile spikes number has a significant effect on increasing grain yield. The above results were consistent with the results obtained in this study on wild wheat. Therefore, it can be stated that the identification of important and effective traits on the yield of wild wheat ancestors is a desirable criterion for selection in crop wheat. In another study on promising wheat lines (Moosavi *et al.*, 2016), harvest index and biomass per plant showed the most direct effect on grain yield. Also, the plant biomass trait showed the most indirect negative effect on grain yield. This result indicates that genotypes that devote a large part of their aerial part assimilates to grain formation and grain filling will have a high grain yield. In general, the plant's efforts to increase the harvest index while increasing water use efficiency are two criteria for the success of a genotype under drought stress that can be used as a final breeding strategy.

In general, the results of our research also confirmed the above discussion. In fact, according to the results, three traits of water use efficiency, seed number per plant, and fertile spikes number per plant lead to increased water use efficiency in the plant. Also, on the other hand, they improve the grain harvest index by increasing the share of assimilated materials transferred to the reproductive parts of the plant. The negative relationship of harvest index with root traits confirms this (Table 3).

In this regard, the wild ancestors of wheat have naturally strengthened this feature and with its help, have adapted for many years under adverse environmental conditions.

In fact, according to biplot results (Figure 1), water use efficiency, fertile spike number per plant, main spike weight, seed weight per spike, seed number per plant, and main stem weight were the most important traits affecting grain yield per plant. Therefore, the first component was called the component of "yield and yield components".

Among the above traits, the important trait of water use efficiency had the highest alignment with increasing grain yield per plant under the conditions of this experiment. Based on the results of this analysis and other statistical analyzes, this trait as an important physiological indicator to select wild germplasm of wheat is recommended under drought stress conditions.

The special features of susceptible ecotype No. 6 were low water use efficiency, high water consumption and allocation of most of the assimilate materials to underground parts, i.e. expansion of root traits instead of increasing yield and yield components. Accordingly, the second component was called the component of the "phenological period". Therefore, it is concluded that excessive water consumption and improper allocation of assimilated materials to underground sections, will reduce the genotype grain yield under stress conditions. In previous studies (Dencic *et al.*, 2000; Moosavi *et al.*, 2013), the efficiency of the principal component analysis method has been reported to identify the desirable traits and genotypes of wheat under water stress. In a study on the genus of *Aegilops* (Moosavi *et al.*, 2017), water use efficiency was suggested as an important criterion for identifying genotypes tolerant to moisture stress conditions.

## Conclusion

Increasing the seed number-related traits per plant is one of the most important strategies for propagation, survival, and adaptation in wild wheat ancestors. It has been used for many years as a natural mechanism for the propagation and survival of wild plants. With this strategy, they

have adapted to adverse environments. Therefore, it is predicted that the election to increase water use efficiency by increasing traits related to the number of seeds per plant, including fertile spike number per plant and seed number per plant, in addition to wild wheat yield, is also effective in improving bread wheat yield. Therefore, plant breeders can use this strategy to increase grain yield in wheat by indirect selection based on these three traits, especially based on the important physiological trait of water use efficiency. Drought-susceptible ecotype 6 had a low grain yield despite high water consumption. This ecotype did not allocate its assimilation well to different parts of the plant. It allocated most of its grown materials to the underground parts. Accordingly, this ecotype had a low grain yield, which was not suitable for drought stress conditions. However, Ecotype 5 with high grain yield and high water use efficiency was less exposed to drought stress and showed a high yield in drought stress.

In general, due to the wide range of tolerance and susceptibility to drought stress in the present germplasm, the use of this gene source is recommended during future breeding programs.

According to our results, high water use efficiency, improvement of traits related to seed number per plant, and the ratio of allocation of assimilates to aboveground or underground plant parts were the main mechanisms of adaptation of tolerant ecotypes to drought stress. Thus, selection based on these traits will lead to the identifying desirable ecotypes for future wheat breeding programs under drought-stress conditions.

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